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# Relationship between Postpartum Diseases and Success of First Artificial Insemination in Dairy Cattle

Hubungan antara Penyakit Postpartus dengan Keberhasilan Inseminasi Buatan Pertama pada Sapi Perah

Diah Ayu Cikita<sup>1</sup>, Reza Taufiq Subagio<sup>1</sup>, Septiyani<sup>2\*</sup>, Vincentius Mubiarto Setiawan<sup>3</sup>, Dwi Kristanto<sup>4</sup>

<sup>1</sup>Doctor of Veterinary Medicine Program, Faculty of Medicine, Padjadjaran University West Java, Indonesia

<sup>2</sup>Department of Basic Medical Sciences, Faculty of Medicine, Padjadjaran University, West Java, Indonesia

<sup>3</sup>Global Dairi Alami, West Java, Indonesia

<sup>4</sup>Faculty of Veterinary Medicine, Brawijaya University, Malang, East Java, Indonesia \**Corresponding author*, Email: drh.septiyani@unpad.ac.id

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#### Abstrak

Ketosis, mastitis, metritis, dan Left displacement of abomasum (LDA) merupakan penyakit yang sering terjadi pada dua minggu pertama pasca partus dan menyebabkan kerugian ekonomi yang besar. Penelitian ini bertujuan untuk menganalisis hubungan penyakit metabolik pada periode postpartus terhadap keberhasilan inseminasi buatan. Penelitian ini dilakukan dengan menggunakan pendekatan observasional cross-sectional. Sebanyak 341 sampel data diperoleh, yang terdiri dari 103 sapi tanpa penyakit, 107 sapi terdiagnosis ketosis, 51 sapi terdiagnosis metritis, 5 ekor sapi terdiagnosis LDA, 20 ekor sapi terdiagnosis kombinasi ketosis dan mastitis, 20 ekor sapi terdiagnosis kombinasi ketosis dan metritis, dan 6 ekor sapi terdiagnosis kombinasi mastitis dan metritis. Analisis data dilakukan dengan menggunakan berupauji chi-square. Nilai P untuk ketosis, mastitis, metritis, LDA, ketosis-mastitis, ketosis-metritis, dan mastitis-metritis masing-masing adalah 0,756., 0,099., 0,972., 0,261., 0,276., 0,276., dan 0,450., sementara itu nilai OR untuk ketosis, mastitis, metritis, ketosis-mastitis, ketosis-metritis, dan mastitis-metritis masing-masing adalah 0,898., 0,424., 1,019., 0,434., 0.434 dan 1.952. Berdasarkan penelitian ini diketahui bahwa sapi perah postpartus yang tidak memiliki riwayat penyakit metabolik memilki peluang keberhasilan inseminasi buatan pertama lebih tinggi, namun secara statisitik menunjukkan bahwa riwayat penyakit pada periode postpartus tidak behubungan signifikan terhadap keberhasilan servis inseminasi buatan pertama pada sapi perah yang di pelihara di peternakan tersebut. Penelitian lebih lanjut untuk menganalisis faktor-faktor yang berhubungan dengan tingkat keberhasilan inseminasi pertama perlu dilakukan dimasa mendatang.

Kata kunci: inseminasi buatan; conception rate; penyakit postpartus

#### Abstract

Ketosis, mastitis, metritis, and *Left displacement of abomasum* (LDA) are diseases that often occur during the first two weeks of lactation and cause significant economic loss. This study aimed to describe the correlation of metabolic diseases during the postpartum period on the success of artificial insemination. This study was conducted using an observational cross-sectional approach. A total of 341 data samples were obtained, which consisted of 103 cattle without disease, 107 cattle diagnosed with ketosis, 51 cattle diagnosed with LDA, 20 cattle diagnosed with a combination of ketosis and mastitis, 20 cattle diagnosed with a combination of ketosis and metritis, and 6 cattle diagnosed with a combination of

mastitis and metritis. Data analysis was carried out using bivariate analysis and tested using the chi-square test. The P values for ketosis, mastitis, metritis, LDA, ketosis-mastitis, ketosis-metritis, and mastitis-metritis were 0.756, 0.099, 0.972, 0.261, 0.276, 0.276, and 0.450, respectively, meanwhile the OR values for ketosis, mastitis, metritis, ketosis-mastitis, ketosis-metritis, and mastitis-metritis are 0.898, 0.424, 1.019, 0.434, 0.434 and 1.952 respectively. Based on this research it is known that postpartum dairy cows do not having a history of metabolic disease has a higher chance of success in the first artificial insemination, but statistically this research shows that a history of disease in the postpartum period does not have a significant effect on the first artificial insemination service in dairy cattle kept on the farm. Further research to analyze factors related to the success rate of first insemination is needed in the future.

Keywords: artificial insemination; conception rate; postpartum disease

### Introduction

Artificial insemination (AI) is a method for inseminating female livestock, both poultry and ruminants, with the help of humans as inseminators (Dako *et al.*, 2022). Artificial insemination allows fertilization to occur without going through the natural mating process using bulls. Several benefits of artificial insemination include improving the genetic quality of livestock because the semen used in AI is obtained from high-quality bulls, and AI may also prevent disease transmission, decreasing costs for raising bulls, increasing the utilization of superior bullsmales, and shortening calving intervals (Setiawan, 2018).

The success of an artificial insemination program can be measured using conception rate parameters. The conception rate (CR) was defined as the percentage of pregnant cows in a female population that were artificially inseminated during the first service (Febriantoro et al., 2015.). Several factors can influence the success of an artificial insemination programme. The main factors that influence the success of AI are the quality of frozen semen, the health status of the female livestock reproductive system, the accuracy and reporting of heat detection, the skills of the inseminator, and other supporting infrastructure and facilities (Pasino et al., 2020). In addition to the health status of the female reproductive system, several other pathological conditions can also interfere with the reproduction of female cattle, which has an impact on the success of artificial insemination. Diseases such as mastitis and ketosis can affect the reproductive performance of cows (Walsh et al., 2007; Wolfenson et al., 2015).

In dairy cattle, the period between 3 weeks before and 3 weeks after parturition is called the transition period, which is one of the most critical physiological stages since most metabolic and infectious diseases occur during this period (Hoseyni et al., 2020; Pascottini et al., 2020; Van Saun, 2016). Major physiological, nutritional, metabolic, and immunological changes occur within this time frame as the production cycle of the cow shifts from a gestational nonlactating state to the onset of copious milk synthesis and secretion (Sordillo & Raphael, 2013). In the transition phase, dairy cows experience dramatic physiological and metabolic changes (Hoseyni et al., 2020). Impaired physiological functions during the transitional phase increase the incidence of metabolic and infectious diseases. Common diseases of dairy cows include retained fetal membranes, metritis, endometritis (as well as cervicitis), mastitis, lameness and abomasal displacement, clinical hypocalcemia, and ketosis. All of these diseases occur most frequently in the early stage of lactation and thus in the period preceding and immediately following insemination (Foditsch et al., 2016; Gilbert and Santos, 2016; Neves et al., 2018). Bisinotto et al. (2012) also reported that metabolic disorders accompanied by poor nutrition during the postpartum period had a negative effect on dairy cows, although this relationship is not known with certainty. Poor management during the transition period was also reported to cause significant economic losses for breeders due to disruption of reproduction and reduced productivity ((Wankhade et al., 2017). Referring to the importance of metabolic disturbances in the postpartum period, the author tries to elaborate on the relationship between metabolic disturbances in the postpartum period and the reproductive status of dairy cows, especially the success of the first insemination.

## **Materials and Method**

This study was conducted from June - July 2023 at a dairy farm owned by PT. Global Dairi Alami (GDA) is located in Subang Regency, West Java. The data used in this study are secondary data consisting of disease history in cows within Days in Milk (DIM) 0-60, history of AI, and pregnancy kit results within a period of 40 days (cows with DIM 100-300 as of 12 July 2023). The inclusion criteria for individuals were as follows: (1) had complete disease history data up to a minimum of DIM 1; (2) had a history of ketosis, mastitis, metritis, or LDA, as well as a combination of these diseases in the fresh period (DIM 0-60); (3) had no history of any disease or disorder during the fresh period (DIM 0-60);and (4) had the results of a pregnancy kit or IB collected more than once. The exclusion criteria for individuals were as follows: (1) had a "not for insemination" status, and (2) had disease or disease combinations for which there was less than 5 cows per category. Based on these criteria, a total of 341 data samples were obtained and subjected to statistical tests. The data consisted of 103 cattle without disease, 107 cattle diagnosed with ketosis, 51 cattle diagnosed with metritis, 5 cattle diagnosed with LDA, 20 cattle diagnosed with a combination of ketosis and mastitis, 20 cattle diagnosed with a combination of ketosis and metritis, and 6 cattle diagnosed with a combination of mastitis and metritis. Both cows and heiffers were included in the data. Cattles with a history of certain diseases or disease combinations during DIM 0-60 were labeled as corresponding diseases. Cattles without a history of disease within 0-60 DIM were considered healthy animals.

Data analysis regarding risk factors was carried out using bivariate analysis with the help of IBM SPSS statistics version 26 software. In this study, the chi-square test was used (Tezera & Ali, 2021). The data were tested using a 95% confidence level with an  $\alpha$  of 5% so that if P < 0.05, the results of the statistical calculations were significant or indicated a significant relationship between the independent variables and the dependent variable.

## **Result and Discussion**

Correlations between ketosis, mastitis, metritis, LDA, and the combination of disease and the first artificial insemination (AI) service were analyzed. These results suggest that disease incidence was not significantly associated with the success of the first AI. There are other factors that may have a greater influence on the success of the first AI.

Table 1 shows that 103 of the 341 individuals (30.2%) were healthy or had no history of fresh disease; 21 of them were pregnant at the first AI, and 82 were not pregnant at the first AI. According to the results of the chi-square test, the p value was 0.275. This value shows a non significant relationship between a history of disease during the fresh period and the success of the first AI. The odds ratio (OR) = 1.391

 Table 1. Correlation between diseases diagnosed during the fresh period and first AI results.

II:	Fi	N	P value		
History	Pregnant	Not pregnant	IN	P value	OR
No fresh diseases diagnosed	21	82	103	0.275	1.391
Diagnosed with fresh diseases	37	201	238		
Total	58	283	341		

Table 2.	Correlation between	ketoses diagnosed	l during the fresh	period and first AI results.
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DIM 0-60 history —	F	First AI		P value	OR
	Pregnant	Not pregnant	IN	r value	OK
Ketosis	20	87	107	0.756	0.898
Healthy	21	82	103		
Total	41	169	210		

(95%CI=0,768-2,520) shows that cows that do not have a history of metabolic disease have a 58,17% chance of success in getting pregnant on the first artificial insemination.

This study (Table 2) showed that a history of ketosis was not significantly associated with the success of the first AI. The OR value = 0.898indicates that cows with a history of ketosis have a 47.31% chance of becoming pregnant at the first insemination. In contrast to our results, Walsh et al. (2007) reported that cows diagnosed with subclinical ketosis within the first two weeks after delivery had a 20% lower conception rate than healthy cows did, while cows diagnosed with clinical ketosis during the same time period had a 50% lower conception rate. A negative energy balance (NEB) was associated with a decrease in the frequency of Gonadotropin hormonereleasing hormone (GnRH) pulses required for Luteinizing Hormone (LH) release, which is necessary to support follicular deviation and ovulation (Matsuyama & Kimura, 2015). NEB conditions can interfere with the sensitivity of the hypothalamus to estradiol- $17\beta$ , causing a decrease in the frequency of the GnRH pulse, which can lead to suppression of the synthesis of LH and FSH (Matsuyama & Kimura, 2015). GnRH is secreted in a pulsed pattern intended to avoid downregulation of the GnRH receptor in the pituitary (Tsutsumi & Webster, 2015). Reduced GnRH pulse act ivity inhibits LH synthesis, resulting in disruption of follicular development and ovulation (Matsuyama & Kimura, 2015).

This study (Table 3) showed that a history of mastitis was not significantly associated with the success of the first AI. The OR value = 0.898 indicates that cows with a history of mastitis have a chance of becoming pregnant at the first insemination of only 29.77%. These results were in accordance with previous study, which described that acute clinical mastitis decreased the conception rate if it occurred mainly 10 days before to 30 days after artificial insemination. Although not as severe as clinical mastitis, subclinical mastitis tends to occur over a long period of time and possibly reduces the conception rate. Mastitis causes an impaired follicular response, including depression of steroid production in the preovulatory follicle, which is associated with a low and delayed preovulatory surge of luteinizing hormone, resulting in delayed ovulation. Clinical and subclinical mastitis also impairs oocvte competency, which is reflected in low blastocyst production (Wolfenson et al., 2015).

This study (Table 4) showed that a history of metritis was not significantly associated with the success of the first AI. The OR values in cases of metritis was 1.019 which shows that cows with a history of this disease have a 50.47% chance of successfully becoming pregnant at the first insemination. Mellado et al. (2018) described that metritis had an effect on the fertility of female cattle. Cows diagnosed with metritis had more service per conception than healthy cows. Armengol & Fraile (2015) compared the conception rates of healthy heifers and cows

DIM 0-60 history —	Fi	First AI			0.0
	Pregnant	Not pregnant	Ν	P value	OR
Mastitis	21	30	51	0.099	0.424
Healthy	21	82	103		
Total	26	128	154		

Table 3. Correlation between mastitis diagnosed during the fresh period and the first AI results.

Table 4. Correlation between metritis diagnosis during the fresh period and the first diagnosis.

DIM 0-60 history —	First AI		Ν	P value	OD
	Pregnant	Not pregnant	IN	P value	OR
Metritis	6	23	29	0.972	1.019
Healthy	21	82	103		
Total	27	105	132		

with those of heifers and cows diagnosed with metritis. The results suggest that metritis caused lower conception in both heifers and cows. The presence of cytological endometritis is one of the major risk factors for anovulation or delayed first ovulation. One mechanism connecting anovulation with uterine disease is the effect of LPS on follicular function. Endotoxin can enter the follicle, and TLR4 is expressed on follicular cells, allowing them to react to LPS and reduce steroidogenesis. Anovulatory postpartum follicles exhibit decreased androstenedione and estradiol synthesis. The association of uterine disease with smaller, slower growing follicles combined with reduced steroidogenesis contributes to prolonged postpartum anovulation, itself a cause of infertility (Santos et al., 2016).

This study (Table 5) showed that a history of LDA was not significantly associated with the success of the first AI. In contrast to our results, Brodzki et al. (2015) explained that a history of LDA caused greater service per conception than did healthy cows. There may be several causes of fertility disorders in cows with abomasal displacement. Impaired uterine involution accompanying LDA is linked to disturbances in mineral and carbohydrate metabolism, which leads to reduced myometrial contractility.

This study (Table 6) revealed that a combination history of ketosis, mastitis, metritis, LDA did not significantly influence the success of the first AI. The OR values in cases of ketosis-

metritis, mastitis-metritis, ketosis-mastitis were 0.434, 1.952 and 0.434 respectively, indicating that cows with a history of these diseases have a chance of successful pregnancy at the first insemination respectively 30.26%, 66.12% and 30.26%. We suggest that there are other risk factors that potentially influence the success of the first AI. Differences in the results between studies may be related to differences in livestock management and practices, animal characteristics, environmental conditions, such as weather or climate, or other factors.

One of the factors that influences the success of AI in the first service is exposure to heat stress. According to a study by Schuller et al. (2014), an increase in the temperature-humidity index causes a significant impact on the conception rate. The negative effects of heat stress on reproductive performance include reduced LH secretion, which is associated with reduced follicular estradiol secretion and reflects impaired dominance of the preovulatory follicle. Heat stress may also impair oocyte and embryo development, thus reducing cow fertility (Wolfenson & Roth, 2019). Oxidative stress was also suggested to be involved in hyperthermia-disrupted fertility.

Another factor that can affect the conception rate is the length of time the placenta is released. Research by Howladder *et al.*, (2019) showed that the percentage of conception will decrease with the length of time it takes for the placenta

 Table 5.
 Correlation between LDA diagnosed during the fresh period and first AI results.

DIM 0-60 History -	First AI		N	D1	OB
	Pregnant	Not pregnant	Ν	P value	OR
LDA	0	5	5	0.261	-
Healthy	21	82	103		
Total	21	87	108		

 Table 6. Correlation between ketosis combined with mastitis diagnosed during the fresh period and the first AI results.

DDIM 0-60 History	First AI		N	P value	
	Pregnant	Not pregnant	Ν	P value	OR
KETOSIS-METRITIS	2	18	20	0.276	0.434
Mastitis-metritis	2	4	6	0.450	1.952
Ketosis-mastitis	2	18	20	0.276	0.434
Healthy	21	82	103		

to release. Kamel et al. (2022) reported that placental retention had a significant effect on the number of services per conception (P<0,001). A decreased rumen pH ( $\leq$ 5.8) was also known to reduce the conception rate at the first AI or the conception rate. Cattle with a rumen pH  $\leq$ 5.8 were at risk of experiencing subacute ruminal acidosis (Ichaisri et al., 2012). A low rumen pH can cause the death and lysis of gram-negative bacteria in the rumen, thereby increasing the abundance of lipopolysaccharide, which can trigger an increase in systemic inflammatory markers, such as the acute phase protein serum amyloid A, cytokines, and haptoglobin, which can exceed physiological levels (Zhao et al., 2018). Lameness in cows may cause stress and an inflammatory response or secondary effect of rumen acidosis, resulting in impaired function of the hypothalamus-pituitary-ovarian axis and adrenal axis, which leads to suppressed reproductive performance (Tsousis et al., 2022; Praxitelous et al., 2023). The release of adrenocorticotropic hormone due to stress caused by laminitis triggers an inhibition of LH release, so that the follicles become persistent and ovulation is delayed (Melendez et al., 2018) (A lower BCS at the first AI is a risk factor that limits the conception rate of the first AI. Cows with a BCS <3.0 had a lower probability of conception at the first AI than cows with a BCS  $\geq$ 3.0. Cows with a BCS  $\leq$ 2.25 were less likely to become pregnant during the first service than cows with a BCS  $\geq$  3.25. This finding suggested that nutritional status at the time of AI is very important for the success of the first AI and that excessive loss of BCS at the start of lactation must be restored before performing AI. Loss of BCS reflects a postpartum energy deficit during early lactation that can interfere with subsequent reproductive performance (Kim & Jeong, 2019).

Technical factors such as the timing of artificial insemination can also affect the success of insemination. Howladder et al. (2019) described that the timing of insemination to the onset of estrus had a significant effect on the percentage of conception. This study showed that the highest percentage of conception was achieved in the 9.1-12, 12.1-14, and 15.1-18 hours after the onset of oestrus, with conception rates of 78.06%, 80.36%, and 76.61%,

respectively. Moreover, the percentage of individuals who conceived after insemination occurred less than three hours after the onset of estrus was only 65%, whereas it was only 69.26% when insemination was performed more than 18 hours after the onset of estrus. This study was based on one company; therefore, the prevalence of bias in the data may be limited by the varied management conditions.

### Conclusion

In conclusion, postpartum dairy cows do not having a history of metabolic disease has a higher chance of success in the first artificial insemination, but statistically this research shows that a history of disease in the postpartum period does not have a significant effect on the first artificial insemination service in dairy cattle. It is possible that there are other risk factors that influence the success of the first AI, such as BCS, rumen pH, heat stress, lameness, timing of the AI, and time of delivery of the placenta. Further research can be carried out regarding the broad range of risk factors for the first AI to determine the strategy to minimize the failure of the first artificial insemination.

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